

AMENDMENTS TO THE CLAIMS:

Claim 1 (Original). An expression vector, comprising a polynucleotide encoding a fusion protein comprising the signal sequence of the gac gene of *Pseudomonas diminuta* and a polypeptide of interest other than gac of *Pseudomonas diminuta*, wherein said signal sequence and said polypeptide of interest are linked in such a way that upon expression of the polynucleotide in a suitable host cell the signal sequence is cleaved off the fusion protein and the polypeptide of interest is released into the periplasm of the host cell.

Claim 2 (Original). The vector according to claim 1, wherein said vector is a plasmid.

Claim 3 (Original). The vector according to claim 1, wherein said vector is a high copy plasmid.

Claim 4 (Original). The vector according to claim 1, wherein the polypeptide of interest is interferon alpha 2.

Claim 5 (Original). The vector according to claim 4, wherein the interferon alpha 2 is selected from the group consisting of interferon alpha 2A and interferon alpha 2B.

Claim 6 (Original). The vector according to claim 1, wherein said signal sequence of the gac gene of *Pseudomonas diminuta* comprises the amino acid sequence (SEQ ID NO 2)

MLRVLHRAASALVMATVIGLAPAVAFA.

Claim 7 (Currently Amended). The vector according to claim 6, wherein said vector further comprises a polynucleotide comprising the promoter region and the ribosomal binding site of the gac gene of *Pseudomonas diminuta*, ~~which~~ said polynucleotide being operatively linked to the polynucleotide encoding the fusion protein comprising the signal sequence and the polypeptide of interest.

Claim 8 (Original). The vector according to claim 7, wherein said polynucleotide comprising the promoter region and the ribosomal binding site comprises the nucleotide sequence (SEQ ID NO 5)

5'-ATCCTGGTTCGTACGCGCCGCCTACAAGTGGTGATCTAGGGGAACGTTCCGGGG

GCGTCGCTGCAACGGCGTCTCCGGATCTGGGTGAGAGGGGAAATCC-3'.

Claim 9 (Original). The vector according to claim 8, wherein said polynucleotide comprising the promoter region and the ribosomal binding site comprises the nucleotide sequence (SEQ ID NO 6)

5'-TCTAGACCAACAACATCTTCAACGTCTACCCGACCAAGATTCAGGAGCCGTCGG
CCGACCTGGGCAATGGGATGTACAGCGGGCTTGCGCCGTTTCGGCTTCACCGGCGG
ATCCTGGTTCGTACGCGCCGCCTACAAGTGGTGATCTAGGGGAACGTTCCGGGGGC
GTCGCTGCAACGGCGTCTCCGGATCTGGGTGAGAGGGGAAATCC-3'.

Claim 10 (Original). A prokaryotic host cell transformed with an expression vector which is compatible with the host cell, said vector comprising a polynucleotide encoding a fusion protein comprising the signal sequence of the gac gene of *Pseudomonas diminuta* and a polypeptide of interest other than gac of *Pseudomonas diminuta*, wherein said signal sequence and said polypeptide of interest are linked in such a way that upon expression of the polynucleotide in a suitable host cell the signal sequence is cleaved off the fusion protein and the polypeptide of interest is released into the periplasm of the host cell.

Claim 11 (Original). The host cell according to claim 10, wherein said vector is a plasmid.

Claim 12 (Original). The host cell according to claim 10, wherein said vector is a high copy plasmid.

Claim 13 (Original). The vector according to claim 10, wherein the polypeptide of interest is interferon alpha 2.

Claim 14 (Original). The vector according to claim 13, wherein the interferon alpha 2 is selected from the group consisting of interferon alpha 2A and interferon alpha 2B

Claim 15 (Original). The host cell according to claim 10, wherein said signal sequence of the gac gene of *Pseudomonas diminuta* comprises the amino acid sequence (SEQ ID NO 2)

MLRVLHRAASALVMATVIGLAPAVAFA.

Claim 16 (Currently Amended). The host cell according to claim 10, wherein said vector further comprises a polynucleotide comprising the promoter region and the ribosomal binding site of the *gac* gene of *Pseudomonas diminuta*, ~~which~~ said polynucleotide being operatively linked to the polynucleotide encoding the fusion protein comprising the signal sequence and the polypeptide of interest.

Claim 17 (Currently Amended). The host cell according to claim 16, wherein said polynucleotide comprising the promoter region and the ribosomal binding site comprises the nucleotide sequence (SEQ ID NO 5)

5'-ATCCTGGTTCGTACGCGCCGCCTACAAGTGGTGATCTAGGGGAACGTTCCGGGG
GCGTCGCTGCAACGGCGTCTCCGGATCTGGGTGAGAGGGGAAATCC-3'

Claim 18 (Currently Amended). The host cell according to claim 16, wherein said polynucleotide comprising the promoter region and the ribosomal binding site comprises the nucleotide sequence (SEQ ID NO6)

5'-TCTAGACCAACAACATCTTCAACGTCTACCCGACCAAGATTCAGGAGCCGTCGG
CCGACCTGGGCAATGGGATGTACAGCGGGCTTGCGCCGTTCCGGCTTCACCGGCCG
ATCCTGGTTCGTACGCGCCGCCTACAAGTGGTGATCTAGGGGAACGTTCCGGGGGC
GTCGCTGCAACGGCGTCTCCGGATCTGGGTGAGAGGGGAAATCC-3'

Claim 19 (Original). The host cell according to claim 10, wherein said host cell is an *E. coli* cell.

Claim 20 (Original). A process for production of a polypeptide of interest, comprising (I) providing a prokaryotic host cell transformed with an expression vector which is compatible with the host cell, said vector comprising a polynucleotide encoding a fusion protein comprising the signal sequence of the *gac* gene of *Pseudomonas diminuta* and a polypeptide of interest other than *gac* of *Pseudomonas diminuta*, wherein said signal sequence and said polypeptide of interest are linked in such a way that upon expression of the polynucleotide in a suitable host cell the signal sequence is cleaved off the fusion protein and the polypeptide of interest is released into the periplasm of the host cell, and

(ii) culturing the prokaryotic host cell under conditions which cause expression of the polynucleotide whereby upon formation of the fusion protein the signal sequence is cleaved off the fusion protein and the polypeptide of interest is released into the periplasm of the host cell.

Claim 21 (Original). The process according to claim 20, further comprising isolation of the polypeptide of interest.

Claim 22 (Original). The process according to claim 20, wherein said vector is a plasmid.

Claim 23 (Original). The process according to claim 20, wherein said vector is a high copy plasmid.

Claim 24 (Original). The vector according to claim 20, wherein the polypeptide of interest is interferon alpha 2.

Claim 25 (Original). The vector according to claim 24, wherein the interferon alpha 2 is selected from the group consisting of interferon alpha 2A and interferon alpha 2B.

Claim 26 (Original). The process according to claim 20, wherein said signal sequence of the gac gene of *Pseudomonas diminuta* comprises the amino acid sequence (SEQ ID NO 2)

MLRVLHRAASALVMATVIGLAPAVAFA.

Claim 27 (Currently Amended). The process according to claim 20, wherein said vector further comprises a polynucleotide comprising the promoter region and the ribosomal binding site of the gac gene of *Pseudomonas diminuta*, ~~which~~ said polynucleotide being operatively linked to the polynucleotide encoding the fusion protein comprising the signal sequence and the polypeptide of interest.

Claim 28 (Currently Amended). The process according to claim 27, wherein said polynucleotide comprising the promoter region and the ribosomal binding site comprises the nucleotide sequence (SEQ ID NO 5)

5'-ATCCTGGTTCGTACGCGCCGCCTACAAGTGGTGATCTAGGGGAACGTTCCGGGG
GCGTCGCTGCAACGGCGTCTCCGGATCTGGGTGAGAGGGGAAATCC-3'.

Claim 29 (Currently Amended). The process according to claim 27, wherein said polynucleotide comprising the promoter region and the ribosomal binding site comprises the nucleotide sequence (SEQ ID NO 6)

5'-TCTAGACCAACAACATCTTCAACGTCTACCCGACCAAGATTCAGGAGCCGTCGG
CCGACCTGGGCAATGGGATGTACAGCGGGCTTGCGCCGTTTCGGCTTCACCGGCGG
ATCCTGGTTCGTACGCGCCGCCTACAAGTGGTGATCTAGGGGAACGTTCCGGGGGC
GTCGCTGCAACGGCGTCTCCGGATCTGGGTGAGAGGGGAAATCC-3'

Claim 30 (Original). The process according to claim 20, wherein said host cell is an E. coli cell.

Claim 31 (Original). The process according to claim 20, said culturing being performed as a multi-stage fermentation process comprising a shake-flask step, optionally a pre-culture step, and a main-culture step.

Claim 32 (Original). The process according to claim 31, wherein said culturing of the procaryotic host cell in the main culture step is performed in a culture medium comprising a substrate for more than about 90% of the cultivation time at a substrate concentration lower than the saturation constant of the substrate, accompanied by high levels of dissolved oxygen concentration, and further accompanied by a steadily decreasing specific growth rate of the bacterial host cells, the process being performed at a temperature which is lower than the optimum temperature for growth of the host cell.

Claim 33 (Original). The process according to claim 32, wherein the concentration of dissolved oxygen in the main culture step is from about 40 % up to about 100% of saturation.

Claim 34 (Original). The process according to claim 32, wherein the steadily decreasing growth rate in the main culture step is from about 2 h⁻¹ to about 0.001 h⁻¹.

Claim 35 (Original). The process according to claim 32, wherein the temperature in the main culture step is between about 22°C and about 35°C.

Claim 36 (Original). The process according to claim 35, wherein the temperature in the main culture step is between about 25°C and about 31°C.

Claim 37 (Original). The process according to claim 36, wherein the temperature in the

main culture step is about 28°C.

Claim 38 (Original). The process according to claim 32, wherein said process is performed at a pH value in the range of about 6.7 to about 7.3 in the pre-culture step and/or the main-culture step.

Claim 39 (Original). The process as claimed in claim 32, wherein the substrate is a carbohydrate or glycerol.

Claim 40 (Original). The process according to claim 39, wherein the carbohydrate is glucose.

Claim 41 (Original). The process according to claim 32, wherein the host cell is an E. coli cell.